INTERNATIONAL JOURNAL OF AGRICULTURE & BIOLOGY
ISSN Print: 1560–8530; ISSN Online: 1814–9596
19–0605/2019/22–4–821–826
DOI: 10.17957/IJAB/15.1135
http://www.fspublishers.org

Full Length Article

Bioremediation of Textile Wastewater through Floating Treatment Wetland System

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Abstract

Textile dye enriched effluents are one of the worst polluters of our water and environmental bodies. Release of these effluents without treatment is not only aesthetically unpleasant but also has damaging impact on flora and fauna. Biological ways of effluents treatment such as floating treatment wetlands (FTWs) are economical, chemical free and environmental friendly. Therefore, this lab study was designed to develop a FTWs system by vegetating two free-floating aquatic plants "*Eichhornia crassipes* (Mart.) Solms and *Pistia stratiotes* L. for the remediation of dye enriched textile wastewater. The efficacy of the system was amplified by inoculating two pollutants degrading and plant growth promoting bacteria "*Bacillus cereus* and *B. subtilis*" as well. A significant decrease, 16.23% in potential hydrogen (pH), 35.27% in electrical conductivity (EC), 54.69% in total dissolved solids (TDS), 45.32% in total suspended solids (TSS), 57.31% in biological oxygen demand (BOD), 66.82% in chemical oxygen demand (COD) and 56.57% in color concentration percentage (CC%) was noted after 3 days period. The quality of treated water matched the standards set by National Environmental Quality Standards (N.E.Q.S.) of Pakistan and Zero Discharge of Hazardous Chemicals (Z.D.H.C.) for municipal and industrial wastewater, thus, suggesting its safe disposal to environmental sink. In conclusion, bio-augmented FTWs could be a promising approach for textile wastewater treatment. © 2019 Friends Science Publishers

Keywords: Textile effluents treatment; Floating treatment wetlands; Plant-bacteria synergism; Hydraulic retention time

Introduction

Textile dyeing and processing industry comprises most environmental unfriendly processes. Many kinds of organic and inorganic chemical compounds along with bases, salts, acids, detergents, bleaching and finishing agents are used in these processes (Tara *et al*., 2019a). Moreover, the presence of color in textile wastewater is one of the major problems. This stops sun light to penetrate in water that directly upsets the photosynthetic activity and reduces oxygen concentration in water. This is highly dangerous to aquatic life (Kadam *et al*., 2018). Moreover, this colored water is heavily polluted with toxicants that are very dangerous to all life forms when ejected untreated to nearby water bodies. Ultimately the rivers not far from the industrial zones are going to be seen dirty and polluted. This is resulting in the decline of their water quality, making it unsuitable for irrigation or other consumption (Charoenlarp *et al*., 2016). Hence, wastewater generated by textile industry must be treated to a suitable quality prior to its discharge to the public water sources.

Conventionally applied techniques like chemical coagulation/flocculation, oxidation, membrane filtration *etc*., for textile effluents treatment, are very expensive. They have high operational and maintenance cost and are source of generating hazardous sludge that requires additional efforts for its safe disposal (Priya and Selvan, 2017). Therefore, the efforts are being made for the use of natural approaches like bioremediation to treat textile wastewater as they are eco-friendly and effective source of treatment. In comparison to existing costly and labor-intensive methods bioremediation is an efficient, environmental pleasant and cost-effective approach for wastewater treatment. It involves plants, bacteria and their synergism for treatment of polluted water, soil and air (Rehman *et al*., 2018). In this system bacteria have catabolic enzymes that degrade hydrocarbons present in contaminated water and soil that supports plant growth in these bodies (Saleem, 2016). On the other hand, plants provide residency, metabolites and nutrients to these bacteria. Plants also take up pollutants, provide filtration media, accumulate organic pollutants and slow down the

To cite this paper: Tusief, M.Q., M.H. Malik, H.N. Asghar, M. Mohsin and N. Mahmood, 2019. Bioremediation of textile wastewater through floating treatment wetland system. *Intl. J. Agric. Biol*., 22: 821–826

flow of contaminants in water through their roots, shoots and leaves. All these mechanisms result in the remediation of polluted water in the form of phytoextraction, rhizofiltration, phytoaccumulation and phytostabilization (Melania *et al*., 2014).

One of the bioremediation approaches is floating treatment wetlands (FTWs), which is soil-less plant technology recently captured space in the field of wastewater treatment. FTWs are the man-made ecosystem that mimics with natural wetlands. It has attracted considerable attention for wastewater treatment for the last few years due to having very simple structure; low operational cost and less labor administrative (Hartshorn *et al*., 2016). In this system plants float on the surface of water providing aesthetic beauty to the location while their roots hang down into the water column to take part in remediation of wastewater by reducing water flow, trapping of suspended particles, increasing sedimentation and providing habitat for growth of microbial communities (Benvenuti *et al*., 2018). In the past few years, FTWs are gaining prominence as a variant of most conventionally used constructed wetlands (CW) for treatment of wastewater of many kinds (Pavlineri *et al*., 2017) owing to have more simple structure and capacity to be applied in any kind of water body irrespective of water depth and fluctuation. Plants are the main component of FTWs system. They perform as catalysts for purifications processes along with assimilation of contaminants directly into their tissues. In addition to this, many of these plants are bioaccumulators (Hegazy *et al*., 2011; Eid *et al*., 2012).

Currently, FTWs have been implemented to treat various kinds of wastewaters *e.g*., municipal, sewage and industrial (Ijaz *et al*., 2016; Ali, 2017). Recently, plant-microbe synergism has been exploited in FTWs for remediation of oil field (Rehman *et al*., 2018) and general textile wastewater (Tara *et al*., 2019b). But in all these studies emergent plants were used for phytoremediation. However, the use of free-floating aquatic plants in interaction with pollutant degrading and plant growth promoting bacteria to treat textile dye enriched effluents has not been practiced to date. Hence, this study was designed to evaluate the potential of free-floating aquatic plants in synergy to their growth promoting and pollutant degrading bacteria for the remediation of textile effluents.

Materials and Methods

Collection and Quality Analysis of Textile Wastewater

The untreated textile wastewater was collected from the outlet of Kamal limited, a famous textile dyeing and processing industry at Khurrianwala, Faisalabad, Pakistan, and transported to the experimental site. The wastewater was stored in a tank, prior to dose into the developed FTWs system. The wastewater was characterized for various physicochemical parameters like pH, EC, TDS, TSS, BOD, COD, color concentration and heavy metals like cadmium (Cd), Chromium (Cr), Nickel (Ni), Lead (Pb) *etc*. (Table 5) by applying standard methods (APHA, 2005) and compared with the wastewater quality standards set by National Environmental Quality Standards (N.E.Q.S.) of Pakistan and Zero Discharge of Hazardous Materials (Z.D.H.C.).

Plants Selection and Collection

For FTWs based phytoremediation process, the plants having dense root system and immunity to survive in local climatic conditions are preferable. "*Eichhornia crassipes* and *Pistia stratiotes"* plants were selected for this study. These plants are abundantly available in waste and fresh water bodies around Faisalabad and Lahore (The textile industry hub of Pakistan). Additionally, the credibility of these plants to treat many types of wastewaters (Aquaculture wastewater, polluted water bodies, RO-Brine solution) has been recognized in previous research studies (Akinbile and Yusoff, 2012; Varghese and Jacob, 2016; Abinaya *et al*., 2018).

Bacterial Strains Collection

Two pollutants degrading and plant growth promoting bacteria of class "*Bacillus*", *B. cereus* and *B. subtilis*, were collected from Soil and Microbiological Lab, Institute of Soil and Environmental Sciences, University of Agriculture, Faisalabad, Pakistan. There they were isolated (Rafique *et al*., 2015) by applying dilution plate technique with general purpose agar media (glucose peptone agar media). Media had composition "1.5 mL/L Glucose, 0.5 mL/L Ammonium sulphate, 0.5 mL/L Potassium hydrogen phosphate, 0.5 mL/L Peptone, 0.1 mL/L Magnesium sulphate hepta hydrate and 20 mL/L Agar. The inoculation of media plates with soil solution was carried out, and then these plates were incubated at 28 ± 2°C for 72 h. The colony farming units (CFU) per gram soil from each soil sample was measured. After isolation these strains were analyzed for their plant growth promotion capabilities by testing their ACC-deaminase activity applying Jacobson's method (Jacobson *et al*., 1994). While their hydrocarbons degrading abilities were tested using Bushnell-Haas broth in 24-well microtiter plates (Hanson *et al*., 1993).

Development of FTWs System

FTWs were designed for the treatment of textile effluents by growing selected plants (5 plants of each type having nearly equal mass) in transparent polypropylene containers of 10 L capacity after filling them with collected textile wastewater. For bioaugmentation of the system, the collected bacterial strains were inoculated in these containers. Plant-bacteria interaction was developed by dipping all plants in 500 mL broth of each bacterium for 40 min. Eight treatment reactors, Wastewater + *E. crassipes*, Waste water + *E.*

crassipes + *B. cereus*, Wastewater + *E. crassipes* + *B. subtilis*, Wastewater + *P. stratiotes*, Waste water + *P. stratiotes* + *B. cereus*, Wastewater + *P. stratiotes* + *B. subtilis*, Wastewater + *B. cereus* and Wastewater + *B. subtilis* were developed. For performance evaluation, a control reactor was also designed to make comparison between treated and untreated wastewater.

Sample Testing

Five hundred mL sample from each treatment reactor was taken after 0 h, 24 h, 48 h and 72 h, hydraulic retention times and tested for pH, EC, TDS, TSS, BOD, COD and color concentration, adopting standards test procedures (APHA, 2005).

Statistical Analysis

The collected data were analyzed statistically in order to check overall significance of data while least significance difference (LSD) test was used to compare difference between treatments mean. Experiment was arranged according to completely randomized design (CRD) with three replications. For all statistical applications, S.A.S. program, version STAT 9.1 of S.A.S. Institute (Clark, 2004) was operated.

Results

Physicochemical Characteristics of Treated Textile Wastewater

Different FTWs systems had significant effect on pH and EC of textile wastewater and all the FTWs systems reduced pH and EC values of wastewater against control at 24, 48 and 72 h (Table 1). The pH value of wastewater before treatment *i.e*., for control reactor was found 8.87 that reduced to 7.43 with combined treatment of water by *E. crassipes* and *B. cereus* after 72 h. So this treatment combination significantly reduced the pH of water up to 16.23% (Table 1). The comparison of mean values for EC of treated water indicated highest value of EC (5.67 dS/m) for control at the start of the experiment that reduced 3.67 dS/m for combined treatment of water with *E. crassipes* plant and *B. cereus* bacteria. There was found 35.27% reduction in EC of treated water under this treatment combination for 72 h retention time (Table 1).

Different FTWs systems had significant effect on TDS and TSS of textile wastewater and all the FTWs systems reduced TDS and TSS of wastewater against control at 24, 48 and 72 h (Table 2). The highest TDS (4891 mg/L) was recorded for control reactor before any kind of treatment *i.e*., at 0 h and this value decreased significantly up to 2216 mg/L when the textile wastewater was treated with *E. crassipes* plant in synergy with *B. cereus* bacteria. This combination recorded 54.69% reduction in TDS of water after 72 h treatment time (Table 2). The data regarding TSS value of the water reflected maximum value of TSS (203 mg/L) for control reactor at 0 h retention time. While, the minimum value of TSS (111 mg/L) was found for *E. crassipes* and *B. cereus* combined treatment after 72 h. Under this treatment combination, a significant reduction (45.32%) in TSS of the treated water was observed (Table 2).

The results for the BOD value of wastewater (Table 3) exposed off the maximum mean value of BOD (171 mg/L) at the start of the experiment before any kind of treatment. This value reduced to 73 mg/L under the joint treatment of *E. crassipes* plant and *B. cereus* after 72 h. This treatment combination made 57.31% reduction BOD value of water under treatment (Table 3). Similarly, the value of COD was found maximum (431 mg/L) for untreated water at the start of the experiment (Table 3) which was reduced considerably up to 143 mg/L for *E. crassipes* and *B. cereus* synergized treatment. For this treatment combination there was found 66.82% reduction in COD of the treated water for 72 h retention time (Table 3). The data in respect of color degradation of textile wastewater reflected remarkable reduction in color concentration of water (56.57%) for *E. crassipes* and *B. cereus* combined treatment for 72 h. Maximum value of color concentration (0.099%) was noted for control reactor at 0 h that reduced to 0.062% at 72 h for *E. crassipes-B. cereus* combined treatment (Table 4).

Discussion

Results indicated that *E. crassipes* plant in synergy with *B. cereus* bacteria reduced all the pollutant indicating parameters of textile wastewater after 3 days of treatment. For this treatment combination and time, all quality parameters were found within the limits set by N.E.Q.S. and Z.D.H.C. (Table 5). There was noted maximum reduction in pH and EC of wastewater for this treatment and time (Table 1). The pH of treated wastewater reduced towards neutral point from alkaline side This reduction in pH by plants might be attributed to the production of organic acids such as formic acid, acetic acid *etc*., during the degradation of organic pollutants present in wastewater. This degradation further enhanced by *B. cereus* bacteria due to having pollutant degrading abilities as recognized previously (Somasiri *et al*., 2008). Likewise, the reduction in EC by the same combination might be due to the decrease of salts present in wastewater that are up-taken by the plants through their roots (Seenivasan *et al*., 2015). Moreover, addition of bacteria in the system boosted up this function by promoting plant growth as reported earlier (Ugya *et al*., 2015).

The TDS value of the treated wastewater also reduced remarkably for *E. crassipes* and *B. cereus* synergized treatment after 72 h retention time (Table 2). This reduction in TDS is because of the plant-bacteria interaction and is attributed to the dense root system of plants (Abinaya *et al*., 2018) that provided space for the accumulation of pollutants present in water in dissolved form while the bacteria

Table 1: Effects of different FTWs systems on pH and EC of textile wastewater

FTWs systems	pH				EC (dS/m)			
	0 hour	24 hours	48 hours	72 hours	0 hour	24 hours	48 hours	72 hours
Control (Textile wastewater without plant and bacteria)	8.87	8.84^{a}	8.79^{a}	8.73^{a}	5.67	5.61^{a}	5.59^{a}	5.58^{a}
Wastewater + *E. crassipes*	8.87	8.75^{ab}	8.61^{b}	8.53^{b}	5.67	5.13^{b}	4.69^{c}	4.23^{c}
Wastewater + *E. crassipes* + *B. cereus*	8.87	8.29^{e}	7.66^{e}	7.43^{e}	5.67	4.79^{f}	4.01^{f}	3.67^{f}
Wastewater + *E. crassipes* + *B. subtilis*	8.87	8.43^{cd}	8.11^{d}	7.91^{d}	5.67	4.96^{de}	4.37^{e}	3.94^{e}
Wastewater + *P. stratiotes*	8.87	8.79^{ab}	8.67^{b}	8.58^{b}	5.67	5.21^{b}	4.85^{b}	4.52^{b}
Wastewater + *P. stratiotes* + *B. cereus*	8.87	8.37^{de}	8.01^{d}	7.93^{d}	5.67	4.90^{e}	4.31^{e}	3.89^{e}
Wastewater + *P. stratiotes* + *B. subtilis*	8.87	8.51^{c}	8.23^{c}	8.07^{c}	5.67	5.11^{bc}	4.87^{b}	4.13^{d}
Wastewater + *B. cereus*	8.87	8.71^{b}	8.59^{b}	8.49^{b}	5.67	5.02^{cd}	4.53^{d}	4.19^{cd}
Wastewater + *B. subtilis*	8.87	8.77^{ab}	8.61^{b}	8.57^{b}	5.67	5.17^{b}	4.67^{c}	4.26^{c}
L.S.D. value at $P < 0.05$		0.095	0.119	0.091		0.096	0.093	0.087

Any two values not sharing a letter in common, within a column, differ significantly at $P < 0.05$
FTWs= floating treatment wetlands; EC= electrical conductivity

Table 2: Effects of different FTWs on TDS and TSS of textile wastewater for various times

FTWs systems	TDS (mg/L)				TSS (mg/L)			
	0 hour	24 hours	48 hours	72 hours	0 hour	24 hours	48 hours	72 hours
Control (Textile wastewater without plant and bacteria)	4891	4879^{a}	4861^{a}	4859^{a}	203	194^{a}	189^{a}	181^{a}
Wastewater + *E. crassipes*	4891	4213^{c}	3788^{c}	3589^{c}	203	166^{bc}	153^{b}	149^{bc}
Wastewater + *E. crassipes* + *B. cereus*	4891	3473^{g}	2649^{f}	2216^{f}	203	145^{d}	127^{e}	111^{e}
Wastewater + *E. crassipes* + *B. subtilis*	4891	3605^{f}	2876^{e}	2479^{e}	203	156^{cd}	132^{de}	119^{de}
Wastewater + *P. stratiotes*	4891	4323^{b}	3925^{b}	3715^{b}	203	173^{b}	161^{b}	157^{b}
Wastewater + *P. stratiotes* + *B. cereus*	4891	3667^{f}	2876^{e}	2489^{e}	203	157^{cd}	138^{de}	121^{de}
Wastewater + *P. stratiotes* + *B. subtilis*	4891	3856^{e}	3103^{d}	2691^{d}	203	160^{c}	140^{cd}	125^{d}
Wastewater + *B. cereus*	4891	4113^{d}	3755^{c}	3556^{c}	203	163^{bc}	150^{bc}	145^{c}
Wastewater + *B. subtilis*	4891	4199^{cd}	3789^{c}	3587^{c}	203	165^{bc}	152^{b}	148^{bc}
L.S.D. value at $P < 0.05$		88.25	79.86	97.81		11.82	11.76	11.21

Any two values not sharing a letter in common, within a column, differ significantly at $P < 0.05$
FTWs= floating treatment wetlands; TDS= total dissolved solids; TSS= total suspended solids

Table 3: Effects of different FTWs systems on BOD and COD of textile wastewater

FTWs systems	BOD (mg/L)				COD (mg/L)			
	0 hour	24 hours	48 hours	72 hours	0 hour	24 hours	48 hours	72 hours
Control (Textile wastewater without plant and bacteria)	171	163^{a}	159^{a}	155^{a}	431	411^{a}	402^{a}	392^{a}
Wastewater + *E. crassipes*	171	139^{bc}	121^{b}	119^{bc}	431	347^{ab}	301^{c}	289^{cd}
Wastewater + *E. crassipes* + *B. cereus*	171	111^{d}	81^{d}	73^{f}	431	279^{b}	175^{d}	143^{e}
Wastewater + *E. crassipes* + *B. subtilis*	171	117^{d}	93^{c}	80^{e}	431	289^{ab}	190^{cd}	167^{ed}
Wastewater + *P. stratiotes*	171	141^{b}	125^{b}	121^{b}	431	361^{ab}	317^{bc}	309^{bc}
Wastewater + *P. stratiotes* + *B. cereus*	171	113^{d}	94^{c}	79^{ef}	431	287^{a}	229^{bc}	196^{cd}
Wastewater + *P. stratiotes* + *B. subtilis*	171	115^{d}	97^{c}	81^{e}	431	301^{ab}	241^{bc}	202^{bcd}
Wastewater + *B. cereus*	171	131^{c}	117^{b}	113^{d}	431	321^{a}	271^{bc}	255^{bc}
Wastewater + *B. subtilis*	171	133^{bc}	119^{b}	115^{cd}	431	333^{a}	283^{ab}	269^{ab}
L.S.D. value at $P < 0.05$		8.27	8.70^{a}	6.13		19.77	11.56	14.20

Any two values not sharing a letter in common, within a column, differ significantly at $P < 0.05$
FTWs= floating treatment wetlands; BOD= biological oxygen demand; COD= chemical oxygen demand

addition in the reactor uplifted this process by enhancing plant growth. Similarly, valuable reduction in TSS of treated textile waster under the combined treatment of plant and bacteria (*E. crassipes* and *B. cereus*) was recorded after maximum retention time (Table 2). This reduction in TSS value might be linked with the root structure of plants in FTWs system that provided filtering media for removing suspended particles in wastewater. Furthermore, these roots also created sites for microbial attachment and settled down the suspended particles by reducing their movement. The presence of bacteria increased the plant growth promotion activities along with degrading and mineralizing the pollutants.

In respect of BOD value of treated wastewater, the *E. crassipes* plant based *B. cereus* bacterial augmented FTWs treatment mechanism made an extensive reduction in it (Table 3). This reduction in BOD value might be endorsed towards the increase of oxygen concentration in wastewater by the plants, due to their photosynthetic activities. This resulted in oxidation process in wastewater which degraded the organic pollutants present in it. The presence of bacteria in the system boosted up this degradation process because of the ability of microbes to transform and decompose

Table 4: Effects of different FTWs systems on color concentration of textile wastewater

FTWs systems	Color concentration (%)			
	0 hour	24 hours	48 hours	72 hours
Control (Textile wastewater without plant and bacteria)	0.099	0.096[a]	0.095[a]	0.095[a]
Wastewater + *E. crassipes*	0.099	0.080[bc]	0.074[b]	0.071[b]
Wastewater + *E. crassipes* + *B. cereus*	0.099	0.061[f]	0.046[c]	0.043[d]
Wastewater + *E. crassipes* + *B. subtilis*	0.099	0.064[ef]	0.050[c]	0.047[cd]
Wastewater + *P. stratiotes*	0.099	0.085[b]	0.076[b]	0.073[b]
Wastewater + *P. stratiotes* + *B. cereus*	0.099	0.068[def]	0.053[c]	0.050[cd]
Wastewater + *P. stratiotes* + *B. subtilis*	0.099	0.071[cde]	0.057[c]	0.053[c]
Wastewater + *B. cereus*	0.099	0.074[cd]	0.071[b]	0.067[b]
Wastewater + *B. subtilis*	0.099	0.078[bc]	0.073[b]	0.070[b]
L.S.D. value at $P < 0.05$		0.010	0.012	0.009

Any two values not sharing a letter in common, within a column, differ significantly at $P < 0.05$
FTWs= floating treatment wetlands

Table 5: Physicochemical properties of textile wastewater before and after treatment and their comparison with N.E.Q.S. and Z.D.H.C. industrial and municipal wastewater standards

Textile effluent properties	Textile wastewater		Units	N.E.Q.S. values	Z.D.H.C. values
	Before treatment	After treatment (For T_2 at TM_4)			
pH	8.87 ± 0.026	7.43 ± 0.017	-	6-10	6-9
EC	5.67 ± 0.012	3.67 ± 0.043	dS/m	-	-
TDS	4891 ± 34.104	2216 ± 34.104	mg/L	3500	-
TSS	203 ± 6.439	111 ± 3.486	mg/L	150	30-150
BOD	171 ± 4.636	73 ± 2.312	mg/L	80	30-150
COD	431 ± 8.671	143 ± 2.607	mg/L	150	40-400
Cd	0.39 ± 0.031	0.13 ± 0.019	mg/L	0.1	0.005-0.2
Cr	1.21 ± 0.107	0.29 ± 0.054	mg/L	1	Not present-0.5
Ni	2.81 ± 0.271	0.14 ± 0.023	mg/L	1	0.1-0.3
Pb	1.79 ± 0.173	0.38 ± 0.066	mg/L	0.5	0.1-2

T_2= treatment reactor containing *Eichhornia crassipes* and *Bacillus cereus*, TM_4 = 72 hours retention time while the values in ± are the standard errors; N.E.Q.S.= National Environmental Quality Standards of Pakistan; Z.D.H.C.= Zero Discharge of Hazardous Chemicals; pH= potential hydrogen, EC= electrical conductivity, TDS= total dissolved solids, TSS= total suspended solids, BOD= biological oxygen demand, COD= chemical oxygen demand, Cd= cadmium, Cr= Chromium, Ni= nickel, Pb= lead

organic matter present in wastewater (Ijaz *et al*., 2016).

Likewise, COD value of treated textile wastewater reduced considerably under *E. crassipes*- *B. cereus* combined treatment with increasing retention time (Table 3), which is related to the depletion of CO_2 in wastewater by plants due to their photosynthetic activity that increased dissolved oxygen in water. This created aerobic conditions in wastewater which favored the aerobic bacterial activity to decrease the COD value as reported earlier (Patel and Adhvaryu, 2016). Regarding the color degradation, a noticeable reduction in color concentration of textile wastewater was observed when it was treated with plants (*E. crassipes*) in association with pollutant degrading bacteria (*B. cereus*) at maximum retention time of 3 days (Table 4). This color degradation is related to the plants ability to take up nutrients and other contaminant through phytoextraction, phytovolatilization and rhizodegradation (Lehl *et al*., 2017) mechanisms. Furthermore, the bacteria presence in the media caused reductive breakage of azo bond (-N=N), chromophore group, of the dye molecules present in water and added up the color degradation process in the system (Sing *et al*., 2015).

Conclusion

The FTWs system of bacterial augmented free floating aquatic wetlands proved its efficacy for all plant-bacteria combinations; however, *E. crassipes* and *B. cereus* synergism made substantial reduction in all pollutant indicating physicochemical parameters of textile wastewater after 72 h retention time. The recorded values of these parameters, after treatment, were found within the limits as set by N.E.Q.S. and Z.D.H.C. for industrial and municipal wastewater standards. Hence, this FTWs system established its worth to treat textile wastewater efficiently in simple, less expensive, chemical free, eco-friendly and sustainable way.

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[Received 10 Apr 2019; Accepted 18 May 2019; Published (online) 20 Aug 2019]